

(19) World Intellectual Property  
Organization  
International Bureau



(43) International Publication Date  
3 March 2005 (03.03.2005)

PCT

(10) International Publication Number  
**WO 2005/019449 A2**

(51) International Patent Classification<sup>7</sup>: C12N 15/00

(21) International Application Number:  
PCT/US2004/021451

(22) International Filing Date: 2 July 2004 (02.07.2004)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:  
60/485,073 3 July 2003 (03.07.2003) US

(71) Applicant (for all designated States except US): **BOARD OF TRUSTEES OPERATING MICHIGAN STATE UNIVERSITY** [US/US]; 450 Administration Building, East Lansing, MI 48824-1046 (US).

(72) Inventor; and

(75) Inventor/Applicant (for US only): **ALLISON, Richard, F.** [US/US]; 1211 Plains Road, Leslie, MI 49251 (US).

(74) Agents: **FALCOFF, Monte, L.** et al.; Harness, Dickey & Pierce, P.L.C., P.O. Box 828, Bloomfield Hills, MI 48303 (US).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM,

AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Declaration under Rule 4.17:

— of inventorship (Rule 4.17(iv)) for US only

Published:

— without international search report and to be republished upon receipt of that report

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: **EXPRESSION OF A RECOMBINANT TRANSGENE**

**A. 5'—Promoter— $\alpha$ -coding— $\alpha$ -IRES—3'UTR→3'**

**B. 5'— $\alpha$ -coding— $\alpha$ -IRES—3'UTR→3'**

**C. 5'— $\alpha$ -3'UTR—IRES—coding→3'**

(57) Abstract: A system for expression of a heterologous polypeptide in a transgenic host cell is disclosed. The system is based upon a transgene comprising a eukaryotic promoter operably linked to a DNA sequence comprising, in the 5' to 3' direction, a DNA sequence complementary to a sequence encoding a heterologous polypeptide, a DNA sequence complementary to an internal ribosome entry site, and a DNA sequence corresponding to a 3' untranslated region of a positive strand single-stranded RNA virus. Following introduction of a stimulus, the host cell synthesizes an RNA molecule complementary to a recombinant RNA encoded by the transgene. The stimulus can be a positive strand single-stranded RNA virus or a nucleic acid thereof. Because the complement of the recombinant RNA comprises an internal ribosome entry site and a sequence encoding a heterologous polypeptide, the host cell can synthesize the heterologous polypeptide.



**WO 2005/019449 A2**